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Applicants : Defu Zeng et al. Confirmation No. 3043
Serial No. : 09/844,544 Art Unit: 1644
Filed : April 27, 2001 Examiner: Marianne DiBrino
For : METHODS FOR INHIBITION OF POLYCLONAL B CELL ACTIVATION AND
IMMUNOGLOBULIN CLASS SWITCHING TO PATHOGENIC AUTOANTIBODIES
BY BLOCKING CD1-MEDIATED INTERACTIONS

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION PURSUANT TO 37 CFR §1.132

Sir/Madam:

I, Dr. Samuel Strober, M.D., do hereby declare as follows:

1. I am a Professor of Medicine in the Department of Medicine, Division of Immunology, Stanford University School of Medicine, Stanford, CA. I received my M.D. from Harvard Medical School, Boston, Magna Cum Laude in 1966. I have over thirty years of experience in immunology. My Curriculum Vitae is attached as Appendix A.
2. I am familiar with the prosecution history of the above-identified patent application and the pending obviousness issues.
3. I am submitting this declaration to show that the use of anti-CD1 antibody to treat lupus is not obvious over the prior art cited by the Examiner. The references used by the Examiner include Zeng et al., Subsets of transgenic T cells that recognize CD1 induce or prevent murine lupus: Role of cytokines. J. Exp. Med., 187:525-536, 1998 and Amano et al., CD1 expression defines subsets of follicular and marginal zone B cells in the spleen: β_2m -dependent and independent forms. J. Immunol., 161:1710-1717, 1998. The Examiner indicates that the results of the Zeng et al. publication in combination with the Amano et al. publication makes obvious the invention of the use

of anti-CD1 mAb to treat lupus. Both these publications are the result of experiments conducted in my laboratory at Stanford University.

4. It is well known in the art that a subset of T cells, CD4 helper T cells, interact with MHC Class II molecules on B cells and thereby augment the production of IgM and IgG antibodies. See Swain S.L., *Immunol. Rev.*, 74:129, 1983 and Swain et al., *J. Immunol.* 132:1118-23, 1984. CD4 T cells that are not CD1 reactive T cells account for 95-96% of all CD4 T cells in NZB/W mice and other well studied mouse strains. See Zeng et al., *J. Clin. Inv.* 112:1211-1222, 2003. CD1 reactive T cells, which interact with the MHC-related molecule, CD1, on B-cells account for about 4-5% of CD4 T cells in these mice strains, whereas in the transgenic mice, i.e., the mice used in the experiments of the Zeng et al *J. Exp. Med.*, 187:525-536, 1998 publication, 100% of CD4 T cells are CD1 reactive because of the introduced transgene. See Zeng et al., *J. Clin. Inv.* 112:1211-1222, 2003 and Zeng et al., *J. Exp. Med.*, 187:525-536, 1998.

5. It is widely believed that CD4 helper T cells are responsible for augmenting the production of pathogenic autoantibodies in lupus. In fact, the landmark paper of Wofsy and Seaman teaches that CD4 helper T cells reactive with MHC Class II molecules, rather than the CD1 reactive T cells, interact with B cells to facilitate the production of pathogenic autoantibodies in lupus prone NZB/W mice. See Wofsy and Seaman, Successful treatment of autoimmunity in NZB/NZW F1 mice with monoclonal antibody to L3T4, *J. Exp. Med.*, 161(2):378-91, 1985. This teaching is based on the amelioration of lupus by administration of anti-CD4 mAb that depletes CD4 helper T cells. This report is widely interpreted in the lupus field as showing that the 95-96% of CD4 T cells that interact with MHC class II and that do not interact with CD1 are required for the production of pathogenic autoantibodies; and that the 3 to 4% of CD4⁺ T cells that interact with CD1 are expected to make a minor contribution to autoantibody production. Thus, there was no reason to believe by those versed in the field that the administration of anti-CD1 mAb, that blocked the contribution of a minority of CD1 reactive T cells, would affect autoantibody production and ameliorate lupus. This is because the vast majority of CD4 T cells (non-CD1 reactive T cells) would still be capable of interacting with B cells via MHC Class II molecules to mediate the disease. Overall, as shown in the present patent application, the unexpected result of the anti-CD1 treatment of NZB/W mice is that

the small minority of CD1 reactive T cells in the NZB/W mice are responsible for helping B cells to produce the majority of the pathogenic autoantibody production.

6. In conclusion, the Amano et al. and Zeng et al. publications do not teach about the contribution of CD1 reactive versus MHC Class II reactive CD4 T cells to lupus, because in the transgenic mouse studies described by the cited art, an unnatural 100% of the CD4 T cells are CD1 reactive, and none are MHC Class II reactive T cells.

7. Zeng et al. and Amano et al. show that there are two subsets of transgenic T cells that react with CD1, one that is Th1 biased and that induces lupus, and another that is Th2 biased and that protects against lupus. These reports do not teach about whether CD1 reactive T cells in non-transgenic mice are predominantly biased toward Th1 or Th2 cytokine patterns, and thus whether the CD1 reactive cells would induce or protect against lupus. In other autoimmune diseases, such as EAE and the autoimmune diabetes of NOD mice, activation of CD1 reactive T cells with the glycolipid, alpha galactosyl ceramide, induces a Th2 pattern and ameliorates disease. See Sharif et al., *Nature Medicine*, 7:1057-62, 2001 and Jahng et al., *J. Exp. Med.*, 194:1789-99, 2001. In the NOD mice, Lehuen and her colleagues showed that deficiency of CD1 reactive T cells induced by targeted inactivation of the CD1 gene worsens disease, and an increase in the number of CD1 reactive T cells by insertion of a V α 14 transgene improves disease. See Lehuen et al., *J. Exp. Med.* 188:1831-39, 1998. These results teach against the pathogenic role of CD1 reactive T cells in autoimmune disease, and instead teach that they are beneficial.

8. Singh et al reported that deficiency of NK T cells in NZB/W mice by targeted inactivation on the CD1 gene worsens lupus. This teaches against a pathogenic role of NK T cells in lupus in NZB/W mice. See Singh et al., 2001, *Arth. Rheum. Suppl.*, Vol 44, pp 283. Chan and his colleagues reported that lupus prone MRL/lpr mice that are CD1 deficient develop worse lupus skin disease than wild-type MRL/lpr mice. See Chan et al., *J. Immunol.*, 167:2985-2990, 2001. Thus, these publications teach against a pathogenic role of CD1 reactive T cells in the lupus disease of MRL/lpr mice. Yang et al. have shown that CD1d deficiency exacerbates lupus in another model of lupus. See Yang et al., *Immunoregulatory role of CD1d in the hydrocarbon oil-induced model of lupus nephritis*, *J Immunol.*, 171(4):2142-53, 2003. Yang et al. (2003) have also demonstrated an expansion of NKT cells, i.e., CD1d reactive cells, with α -GalCer and improved dermatitis in a model

used to study the pathogenesis of lupus. See Yang et al., Repeated α -galactosylceramide administration results in expansion of NK T cells and alleviates inflammatory dermatitis in MRL-lpr/lpr mice, J Immunol., 171(8):4439-46, 2003.

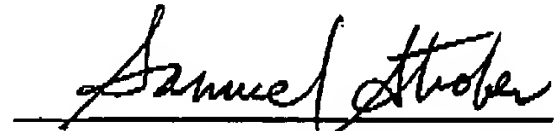
9. When the teaching that activation of CD1 reactive T cells ameliorate autoimmune disease such as EAE and diabetes via their Th2 bias and the teaching that deficiency of CD1 reactive T cells in lupus prone NZB/W and MRL/lpr mice worsens lupus are taken into account, a person of skill in the art would not expect that anti-CD1 mAb treatment would ameliorate lupus. They would expect the opposite.

10. Overall, the Zeng et al. and Amano et al. references do not make obvious the use of anti-CD1 antibody for the treatment of lupus.

11. I declare further that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that making of willful false statements and the like are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful statements may jeopardize the validity of the applications or any patent issuing thereon.

Dated: April 27, 2006

Respectfully submitted,



Samuel Strober, M.D.

Professor of Medicine

Department of Medicine

Division of Immunology

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Stanford, CA

**APPENDIX A
CURRICULUM VITAE**

Samuel Strober, M.D.

Military Status: Three years' active duty in United States Public Health Service, Honorable Discharge, June 23, 1970

Education:

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| 1961 | A.B. Columbia College, New York City, Liberal Arts |
| 1966 | M.D. Harvard Medical School, Boston, Magna Cum Laude |

Honors:

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| 1966 | Leon Resnick Memorial Research Prize, Harvard Medical School |
| 1966 | Alpha Omega Honorary Society |
| 1971-1976 | Career Development Award from NIAID |
| 1984 | Diane Goldstone Memorial Lecturer, Massey Cancer Center |
| 1986 | John Putnam Merrill Memorial Lecturer, Harvard Medical School |
| 1987-1989 | Federal Advisory Committee, Transplantation Biology and Immunology Subcommittee, NIH |
| 1989 | Ray A. and Joan B. Kroc Visiting Professor, University of Michigan |
| 1993 | E. Donnall Thomas Annual Lecture, Fred Hutchinson Cancer Research Center |
| 1996 | President, Clinical Immunology Society |

Training and Experience:

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| 1962-1963 | Research Fellow, part time, Surgical Research Laboratory, Peter Bent Brigham Hospital, Boston, MA, Head: Professor J.E. Murray |
| 1963-1964 | Research Fellow, Cellular Immunology Research Unit, Oxford University, Oxford, England, Head: Professor J.L. Gowans |
| 1965-1966 | Research Fellow, Surgical Research Laboratory, Peter Bent Brigham Hospital, Boston, MA |
| 1966-1967 | Intern, Department of Medicine, Massachusetts General Hospital, Boston, MA, Professor Alexander Leaf, Head, Department of Medicine |
| 1967-1970 | Research Associate, Laboratory of Cell Biology, National Cancer Institute, NIH, Bethesda, MD, Head: Dr. L. W. Law |
| 1970-1971 | Senior Assistant Resident, Department of Medicine, Stanford University School of Medicine, Stanford, CA |
| 1971-1972 | Instructor in Medicine, Department of Medicine, Division of Immunology, Stanford University School of Medicine, Stanford, CA |
| 1972-1978 | Assistant Professor of Medicine, Department of Medicine, Division of Immunology, Stanford University School of Medicine, Stanford, CA |

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| 1976-1981 | Investigator, Howard Hughes Medical Institute, Miami, FL |
| 1978-1982 | Associate Professor of Medicine, Department of Medicine, Division of Immunology, Stanford University School of Medicine, Stanford, CA |
| 1978-1997 | Chief, Division of Immunology and Rheumatology, Department of Medicine, Stanford University School of Medicine, Stanford, CA |
| 1982-present | Professor of Medicine, Department of Medicine, Division of Immunology, Stanford University School of Medicine, Stanford, CA |

Editorial Boards:

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| 1982-1984 | Associate Editor, Journal of Immunology |
| 1982-1985 | Associate Editor, Transplantation |
| 1984-present | Associate Editor, International Journal of Immunotherapy |
| 1992-present | Associate Editor, Transplantation Immunology |
| 1998-2002 | Member, Biology of Blood and Marrow Transplantation |

Institutional Boards:

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| 1992-2005 | Member, Board of Directors, La Jolla Institute for Immunology |
| 2005-Present | Chairman, Board of Directors, La Jolla Institute for Immunology |

Societies:

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| 1989-1997 | Councilor, Clinical Immunology Society |
| 1996 | President, Clinical Immunology Society |
| | American Association of Immunologist |
| | American Society for Clinical Investigation |
| | American College of Rheumatology |
| | American Society of Transplantation Physicians |
| | Western Society for Clinical Investigation |
| | American Association of Physicians |
| 1986-1989 | Councilor, Transplantation Society |

Advisory Committee:

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| 1987-1989 | NIH Transplantation Biology and Immunology Study Section |
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